



VOLUME 2/2005

MICRO NEWS

INTRODUCTION TO ASTM D7102-04:

STANDARD GUIDE FOR DETERMINATION OF ENDOTOXIN ON STERILE MEDICAL GLOVES

Amy Jo Karren, B.S., RM(NRM), CQA
Microbiology Section Leader

Complications from pyrogen on medical gloves prompted action from the FDA to regulate the bacterial endotoxin concentration on sterile medical gloves. In February 2005, ASTM published the Standard Guide for Determination of Endotoxin on Sterile Medical Gloves as the new standard ASTM D 7102-04.

Pyrogens from various sources can cause fever in patients subjected to medical procedures. Often, surgeon's sterile latex gloves were determined to be the source of febrile reactions. Lipopolysaccharides unique to the outermost wall of gram negative bacteria, known as bacterial endotoxin, have very potent proinflammatory effects. These effects can irritate the skin, induce respiratory problems, fever and shock in both the patient and the glove wearer. Pyrogenicity can be amplified with latex interaction. The standard applies to all sterile medical gloves, whether they are latex, polyvinyl or nitrile.

The new endotoxin standard defines sterile medical gloves as a medical device. The packaging of the medical gloves (e.g., one glove or two) determines the definition of the device.

ASTM D7102-04 now requires sterile medical gloves to be analyzed and determined to be non-pyrogenic.

SCOPE OF THE STANDARD:

The guide was established by compiling current regulations on bacterial endotoxin testing and medical gloves. The references include EN455-3:1999 (*Medical Gloves for Single Use – Part 3: Requirements and Testing for Biological Evaluation*) and ANSI/AAMI ST72:2002 (*Bacterial Endotoxins – Test Methodologies, Routine Monitoring and Alternatives to Batch Testing*).

This guide allows the use of the four traditional Limulus Amebocyte Lysate (LAL) test methods (gel-clot, kinetic turbidimetric, kinetic chromogenic, and end-point chromogenic), to determine the presence of bacterial endotoxin on sterile medical gloves. The only pyrogen addressed in this standard is bacterial endotoxin.

A standard sample preparation method is described in this guide. This guide is appropriate for testing final product that is subjected to all processes, including sterilization. Bacteria

(cont. page 2)

IN THIS ISSUE

Endotoxin on Sterile Medical Gloves pages 1-2
 The NEW nelsonlabs.com page 2
 Vmax and The Future of
 Radiation Validation page 3
 Antimicrobial Finishes on Textiles
 and other Products pages 4-5
 USP 797 page 5
 Sterilization Workshop page 6



6280 So. Redwood Road
 Salt Lake City, Utah
 84123-6600-80
 800.826.2088

*Introduction
to ASTM
D7102-04:
Standard
Guide for
Determination
of Endotoxin
on Sterile
Medical Gloves
(cont.)*

REFERENCES

ASTM D7102-04 Standard Guide for Determination of Endotoxin on Sterile Medical Gloves

ANSI/AAMI ST72:2002, Bacterial endotoxins - Test methodologies, routine monitoring, and alternatives to batch testing. Association for the Advancement of Medical Instrumentation, Arlington, VA.

EN455-3:2000. December 1999. British Standard, Medical Gloves for Single Use, Part 3: Requirements and Testing for Biological Evaluation.

United States Pharmacopeia 28 & National Formulary 23. 2005. Bacterial Endotoxins Test <85>, and Transfusion and Infusion Assemblies and Similar Medical Devices <161>. United States Pharmacopeial Convention, Inc., Rockville, MD.

may continue to grow on non-sterile gloves, therefore it is appropriate to analyze sterile medical gloves.

SAMPLING:

The sampling plan should be based on the size of the batch. Three percent of the batch should be tested with a minimum of 3 and a maximum of 10 pairs of gloves per batch. For batch sizes under 30 units, two pairs of gloves may be analyzed. The bacterial endotoxin test should be performed on each batch of gloves with a non-pyrogenic label claim. FDA expects that sterile medical gloves will meet the bacterial endotoxins test criteria limit of 20 endotoxin units per device.

SAMPLE PREPARATION:

An extraction of the gloves is necessary to perform the LAL test. The extraction parameters are well detailed in the guide. The extraction involves immersing the outside surface of the gloves in LAL reagent water. The extraction is performed at 37° to 40° C for 40 to 60 minutes, and must ensure that all exterior surfaces of the gloves that have patient contact, come in contact with the extraction fluid.

Powder and other substances that leach from

the gloves may interfere with the LAL assay. Generally, interference is overcome by simple dilution. The guide also recommends against centrifugation or filtration of particles, as this may remove endotoxin from the test sample.

INTERPRETATION:

Previously, no endotoxin limits had been established for medical gloves. The 20 endotoxin unit per device (20 EU/device) limit was established for medical devices by the United States Pharmacopeia. By meeting this limit a manufacturer can now label their gloves as non-pyrogenic.

The implementation of this standard is beneficial because it minimizes the safety concerns resulting from endotoxin complications. Medical device and pharmaceutical manufacturers also benefit, because the utilization of gloves labeled as non-pyrogenic in the production environment can significantly decrease the probability of endotoxin contamination on devices or pharmaceutical products. Endotoxin transfer is amplified with the use of water, solvents, or detergents. Transfer of endotoxin from gloves to products is a common source of contamination.

The NEW nelsonlabs.com



Nelson Laboratories is proud to announce the creation of our new, client-based, website. Our new site will go live summer 2005. The website name, **www.nelsonlabs.com**, will not be changing; however, the design will be excitingly new and different. You will still be able to find the same valuable information but in a much faster, easier way. For a more pleasant nelsonlabs.com experience we have included the following:

- Online sample submission
- New search engine by:
 - Industry
 - Standard test method
 - Catalog code
 - Key word / Test name
- Request a quote or information
- Access to secure client site
- View pricing, study outline, Nelson contact person for each test on one page

Please feel free to send comments to our marketing team at **marketing@nelsonlabs.com**.



RADIATION VALIDATION FOR A NEW AGE:

VDmax and The Future of Radiation Validation

Martell Winters, B.S. RM/SM(NRM)

The new version of ISO 11137-2 is set to be published in 2006. The new standard, which has been in development for several years, currently includes the VDmax validation procedure. When the new version of ISO 11137-2 is published, nearly every country in the world is expected to accept it. This should put an end to the difficulties of complying with different requirements and standards for different countries.

There was one item of debate related to validating product with very low bioburden. This discussion resulted in a new dose-setting table for Method 1 when the bioburden average is less than 1.0 colony forming unit (CFU) per product. This also resulted in another table for VDmax which validates a 15 kGy sterilization dose in addition to 25 kGy. Thus, when this document is published, companies will be able to choose between a VDmax validation of either 15 kGy (maximum bioburden of 1.5 CFU) or 25 kGy (maximum bioburden of 1,000 CFU).

The Association for the Advancement of Medical Instrumentation (AAMI) recently handed a New Work Item Proposal to the Radiation Working Group to begin writing a document which will provide a wide range of VDmax doses. This new document is expected to include tables for validating sterilization doses of 15 kGy to 35 kGy in 2.5 kGy increments. The new document will likely allow validation of a sterilization dose by either:

- A. Determining the bioburden estimate for the product and choosing the appropriate VDmax sterilization dose.
- B. Choosing the preferred VDmax sterilization dose for the product and ensuring that the bioburden estimate is below the limit established for that dose.

These options should allow the wide range of sterilization doses currently available in Method 1, but with the smaller sample size of the VDmax method (10 samples rather than 100 for sterility tests). The AAMI Radiation Working Group hopes to have the document published some time in 2006. Once this document is published, it will likely make Method 1 close to obsolete.

For additional information, please contact Martell Winters at 801-963-2600 (or 800-826-2088) ext. 9064, or Dr. Wendy Wangsgard at ext. 9030.

Martell is a member of the AAMI Sterilization Standards Committee and participates on the Working Groups of Radiation, Microbiological Methods, Sterility Assurance Level and Terminology. He is also a member of the American Association of Tissue Banks Standards Committee.



In 2006 a new version of AAMI/ISO/EN 11137-2 is slated to be published, and will include the VDmax validation method for both 15 and 25 kGy.

REFERENCES

ANSI/AAMI/ISO 11137-1994 - "Sterilization Of Health Care Products - Requirements For Validation And Routine Control - Radiation Sterilization".
ANSI/AAMI/ISO.



ANTIMICROBIAL FINISHES ON TEXTILES AND OTHER PRODUCTS

*Bryan E. Wilson,
B.S. RM(NRM)
Pharmaceutical Section
Study Director
Disinfectant and
Antimicrobial Studies*

REFERENCES

- AATCC Test Method 147-1998. 2001. Antibacterial Activity Assessment of Textile Materials: Parallel Streak Method. American Association of Textile Chemists and Colorists. Research Triangle Park, NC.
- ASTM E2149-01 Standard Test Method for Determining the Antimicrobial Activity of Immobilized Antimicrobial Agents Under Dynamic Contact Conditions
- Technical Manual of the American Association of Textile Chemists and Colorists (AATCC). AATCC Test Method 100-2004.



INTRODUCTION:

Under suitable conditions, microorganisms which inhabit the soil, water, and air can grow on textile materials. This can result in biodeterioration of the material as well as present health risks to consumers. Application of antimicrobial agents to textile materials as a final finishing process helps to alleviate this problem. Methods for evaluating the effectiveness of antimicrobial finishes on textiles are available and recommended to validate your product.

BACKGROUND:

Biodeterioration has been defined as “any undesirable change in the properties of a material caused by the vital activities of organisms” (H. J. Hueck). Textiles made from natural fibers are generally more susceptible to biodeterioration by microorganisms than the more resistant synthetic man-made fibers. Bacteria and fungi may attack the textile material as a whole, the textile fibers, or a single component such as a plasticizer. Products such as starch, protein derivatives, fats, and oils used in the finishing of textiles can also promote microbial growth.

The growth of microorganisms in the organic soil load on clothing can result in staining, retention of unpleasant odors and even pose potential health risks to consumers. Conditions ranging from mild discomfort and irritation to allergic sensitization and toxic responses may occur. Methods to control biodeterioration and decrease consumer health risks from microbial growth on textiles must include consideration of the beneficial, as well as harmful, organisms to ensure the non-target organisms or natural flora remain unaffected. The adaptation of

microorganisms to the control methods must also be considered.

APPLICATION:

The best method of avoiding microbial deterioration is to use synthetic materials, which have been shown to be inherently resistant to microbes. An alternative strategy is to apply antimicrobial chemicals which are normally incorporated into the finished textile product. So far, a preservative agent has yet to be discovered which gives complete protection or is without some disadvantage. The requirements for an “ideal” antimicrobial would include:

- Effective against a wide range of microorganisms, particularly bacteria and fungi
- Active during the life of the product
- Non-toxic to humans at the concentrations used
- Colorless and odorless
- Effective at low concentrations
- Inexpensive and easy to apply
- Resistant to sunlight and leaching from the fabric
- Does not adversely affect the strength of the material
- Compatible with water-repellent and flame-retardant agents, dyes, and other textile auxiliaries

Application of antimicrobials to textile fabrics is usually performed as a final finishing process. Antimicrobials are also used to provide hygienic finishes for fabrics that will be used in health-care products. These finishes are classified as either renewable or durable, although durable finishes are gradually removed during laundering. Renewable finishes can be replaced during laundering. These include ammonium compounds for certain applications.

EVALUATION:

Methods for evaluation of the effectiveness of an antimicrobial finish have been outlined in the Technical Manual of the American Association of Textile Chemists and Colorists (AATCC), Volume 80, 2005.

AATCC Test Method 100 describes inoculating both treated and untreated swatches of test material with an appropriate test organism. At

time intervals, based on the expected effectiveness of the treated material, the samples are extracted in a neutralizer medium, diluted, and plated accordingly. Test interval times are generally 0, 24, and occasionally, 48 hours or more. Organism recovery from the treated and untreated samples are compared to determine log reduction and percent reduction over time.

AATCC Test Method 147, Parallel Streak Method, is an effective qualitative test for determining the antimicrobial activity of treated textile materials. Five parallel streaks of an appropriate organism are made on an agar surface plate. The streaks are made with a single loop of inoculum, resulting in decreasing growth of the organism from one end of each streak to the other and from one streak to the next and, therefore, increasing degrees of sensitivity. A test sample is placed perpendicular to the streaks on the plate and incubated. After incubation, the plates are examined for the inhibition of growth underneath and near the edge of the test material along each streak of inoculum. The widths of these areas of inhibition are measured and average widths calculated. Both treated and untreated samples should be tested. Although the test is qualitative, a series of test samples treated with different concentrations of antimicrobial can be tested and compared to determine a rough estimate of the lowest concentration needed to inhibit the growth of a particular organism.

The ASTM E2149 Standard Test Method for Determining the Antimicrobial Activity of Immobilized Antimicrobial Agents Under Dynamic Contact Conditions is another means of testing an antimicrobial product. In this

method, a sample is placed in a container with an appropriate test organism. The container is placed on an orbital shaker and incubated. The concentration of the organism is measured both before and after incubation, and a log reduction value is determined. The type of test challenge applied in this method is extreme, and the test is most effective for testing antimicrobials that are "fixed" to the product and do not leach out into the environment.

Other methods with specific application to textiles have been proposed by ASTM, AATCC, and other organizations, all of which follow the same basic techniques previously described. However, testing standards for the evaluation of antimicrobials incorporated into medical devices such as catheters, Luer-loc connectors, stopcocks, and other devices, currently do not exist. As this is a relatively new area in the application of antimicrobials, protocols for these types of tests are evaluated on a case-by-case basis by the FDA. Nelson Laboratories, Inc. has successfully developed many of these protocols for our clients.

For more information regarding the testing of your antimicrobial textile, medical device coating, or other product, please contact Bryan Wilson at 800-963-6280 ext. 9029 or e-mail to bwilson@nelsonlabs.com.



With the publication of USP <797> the requirements for pharmacists performing compounding have increased dramatically. In order to be in compliance, the compounding pharmacist must now address plans for microbiology, sterility, pyrogen and potency testing requirements on compounded sterile preparations (CSPs), as defined in USP <797>.

Nelson Laboratories has established and validated procedures to perform all of the required testing. Our professionally trained personnel and state of the art

**USP
<797>**

testing facility ensure that we can meet the testing needs of the compounding pharmacy. All operations are performed in a GLP/GMP compliant environment that is designed for efficient processing of samples.

If your compounding pharmacy has a microbiology and chemistry laboratory, we would also be able to provide you with consulting and training, in order to easily implement the new requirements. For additional information please contact Amy Karren at akarren@nelsonlabs.com.



ETHYLENE OXIDE & RADIATION STERILIZATION WORKSHOP

TOPICS TO BE PRESENTED:

Ethylene Oxide Sterilization • Radiation Sterilization • Biocompatibility
Routine Tests • Packaging • Sterilization Basics

Location: San Diego, California
Date: August 15-16, 2005 • 9:00 AM-5:00 PM
Price: Before July 15, 2005 \$550.00,
after \$600.00
Contact: Jared Forsyth, jforsyth@nelsonlabs.com
800-963-6280, ext. 9051

For more info: www.nelsonlabs.com/seminars.htm

LIMITED SEATING — DON'T MISS OUT — REGISTER NOW!

*Celebrating 20 Years of
Test Service Excellence*



6280 So. Redwood Road
Salt Lake City, UT 84123-6600-80



PPSR1 1ST CLASS
US Postage
PAID
SLC, UT 841
Permit No. 7259